

Matter: 26208
Serial #: 10/500,447
Date: April 3, 2006

For transformation, recombinant pBI/tPA and pSK/tPA constructs were constructed through the combination of pBI121 with tPA from human. The process for making the constructs are as follows: the primer No. 3 (sense) was designed to have Xba I restriction site fused to the start codon of tPA nucleotide sequence, 5'-AATCTAGACATGGATGCAATGAAGA-3' (SEQ ID NO. 3) and the primer No. 4 (antisense) was designed to contain the stop codon followed by SacI site, 5'-ATGATCTCTGGTCACGGTCGCATGTT-3' (SEQ ID NO. 4); From pETPFR (ATCC #40403) harboring tPA gene, tPA was amplified using tPA specific primers by PCR techniques. Polymerase Chain Reaction (PCR) was performed for 30 cycles of the following reaction: 94°C 30 sec, 53° 30 sec, 72°C 2 min. The resulting 1.7 kb DNA fragment (SEQ. No. 6) was ligated to pT7BlueR (Novagen) plasmid to obtain the recombinant pT/tPA. From it, the tPA DNA was excised by cutting with XbaI and SacI to be fused to pBluescript II SK (Stratagene) and to GUS-DNA deleted pBI121 (Clonetech) by digesting with XbaI and SacI to obtain pSK/tPA and pBI/tPA (not shown). After this, two kinds of recombinant plasmid were introduced into *E. coli* DH5 α and *E. coli* XL-1 Blue. And then, it was identified that the sequence inserted in the resulted 1.7 kb DNA fragment was tPA full length sequence.

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In the Sequence Listing:

Please amend the sequence listing by replacing the 3 pages of Sequence Listing as originally filed with the substitute paper copy and substitute computer readable copy of the sequence listing attached hereto.